

# SEQUENCE LISTING

<110> E.I. duPont de Nemours and Company  
Genencor International, Inc.  
Cervin, Maggie  
Soucaille , Philippe  
Valle, Fernando

<120> PROCESS FOR THE BIOLOGICAL PRODUCTION OF 1,3-PROPANEDIOL WITH HIGH YIELD

<130> CL2180 US NA

<150> 60/416192  
<151> 2002-10-04

<160> 68

<170> PatentIn version 3.1

<210> 1

<211> 1137

<212> DNA

<213> Artificial Sequence

<220>

<223> partial DNA sequence of plasmid pLoxCat27 comprising the LoxP-Cat  
-LoxP cassette

<400> 1  
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gtatagtata cattatacga agttatctag agttgcatgc ctgcaggtcc gaatttctgc 120  
cattcatccg cttattatca cttattcagg cgtagcacca ggcgtttaag ggcaccaata 180  
actgccttaa aaaaattacg ccccgccctg ccactcatcg cagtactgtt gtaattcatt 240  
aagcattctg ccgacatgga agccatcaca aacggcatga tgaacctgaa tcgccagcgg 300  
catcagcacc ttgtgcctt gcgtataata tttgcccatt gtgaaaacgg gggcgaagaa 360  
gttggtccata ttggccacgt ttaaataaaa actggtgaaa ctacccagg gattggctga 420  
gacgaaaaac atattctcaa taaacccttt agggaaatag gccaggtttt caccgtaaca 480  
cgccacatct tgcgaatata tgtgtagaaa ctgccggaaa tcgtcgtggt attcactcca 540

gagcgatgaa aacgtttcag tttgctcatg gaaaacggtg taacaagggt gaacactatc	600
ccatatcacc agctcaccgt ctttcattgc catacgggaat tccggatgag cattcatcag	660
gcgggcaaga atgtgaataa aggccggata aaacttgtgc ttatTTTTtct ttacggtcct	720
taaaaaggcc gtaatatcca gctgaacggt ctggttatag gtacattgag caactgactg	780
aaatgcctca aaatgttctt tacgatgcc a ttgggatata tcaacggtgg tatatccagt	840
gattTTTTtcc tccatttttag cttccttagc tcctgaaaat ctcgataact caaaaaatac	900
gcccggtagt gatcttattt cattatggtg aaagttggaa cctcttacgt gccgatcaac	960
gtctcatttt cgccaaaagt tggcccaggg cttcccggta tcaacaggga caccaggatt	1020
tatttattct gcgaagtgat cttccgtcac aggtatttat tcggactcta gataacttcg	1080
tatagtatac attatacgaa gttatgaagg gcgaattctg cagatatcca tcacact	1137

<210> 2

<211> 61

<212> DNA

<213> Artificial Sequence

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<223> Primer ArCA1

<400> 2

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c	61
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<210> 3

<211> 62

<212> DNA

<213> Artificial Sequence

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<223> Primer ArCA2

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ttccagatca ccgcagaagc gataaccttc accgtgaatg gtcatatgaa taccctcctt	60
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ag	62
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<210> 4

<211> 24

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer ArcA3

<400> 4

agttggtaac acgcaacacg caac

24

<210> 5

<211> 23

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer ArcA4

<400> 5

cgcagaagcg ataaccttca ccg

23

<210> 6

<211> 1320

<212> DNA

<213> Artificial Sequence

<220>

<223> Partial sequence of pLoxCat1 comprising the lox-Cat-loxP cassette

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agctggatcc ataacttcgt ataatgtatg ctatacgaag ttatctagag tccgaataaa	120
tacctgtgac ggaagatcac ttcgcagaat aaataaatcc tgggtgtccct gttgataccg	180
ggaagccctg ggccaacttt tggcgaaaat gagacgttga tcggcacgta agaggttcca	240
actttcacca taatgaaata agatcactac cgggcgtatt ttttgagtta tcgagatttt	300
caggagctaa ggaagctaaa atggagaaaa aaatcactgg atataccacc gttgatatat	360
cccaatggca tcgtaaagaa cattttgagg catttcagtc agttgctcaa tgtacctata	420
accagaccgt tcagctggat attacggcct ttttaaagac cgtaaagaaa aataagcaca	480
agtttttatcc ggcctttatt cacattcttg cccgcctgat gaatgctcat ccggaattcc	540
gtatggcaat gaaagacggg gagctggtga tatgggatag tgttcaccct tgttacaccg	600
ttttccatga gcaaactgaa acgttttcat cgctctggag tgaataccac gacgatttcc	660
ggcagtttct acacatatat tcgcaagatg tggcgtgtta cggtgaaaac ctggcctatt	720
tccctaaagg gtttattgag aatatgtttt tcgtctcagc caatccctgg gtgagtttca	780

ccagttttga tttaaacgtg gccaatatgg acaacttctt cgccccggtt ttcacccatgg	840
gcaaataatta tacgcaaggc gacaaggtgc tgatgccgct ggcgattcag gttcatcatg	900
ccgtttgtga tggcttccat gtcggcagaa tgcttaatga attacaacag tactgcatg	960
agtggcaggg cggggcgtaa tttttttaag gcagttattg gtgcccttaa acgcctggtg	1020
ctacgcctga ataagtata ataagcggat gaatggcaga aattcggacc tgcaggcatg	1080
caactctaga taacttcgta taatgtatgc tatacgaagt tatgcggccg ccatatgcat	1140
cctaggccta ttaatatcc ggagtatacg tagccggcta acgttctagc atgcgaaatt	1200
taaagcgctg atatcgatcg cgcgcagatc tgtcatgatg atcattgcaa ttggatccat	1260
atatagggcc cgggggttata attacctcag gtcgacgtcc catggccatt gaattcgtaa	1320

<210> 7

<211> 61

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer GalA

<400> 7

tcggttttca cagttgttac atttcttttc agtaaagtct ggatgcatat ggcggccgca	60
t	61

<210> 8

<211> 65

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer GalP2

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catgatgcc tccaatatgg ttatttttta ttgtgaatta gtctgtttcc tgtgtgaaat	60
tggtta	65

<210> 9

<211> 60

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer GlkA

<400> 9  
acttagtttg cccagcttgc aaaaggcatc gctgcaattg gatgcatatg gcggccgcat 60

<210> 10

<211> 67

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer Glk2

<400> 10  
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attgtta 67

<210> 11

<211> 1270

<212> DNA

<213> Artificial Sequence

<220>

<223> LoxP-cat-loxP Trc cassette "insert"

<400> 11  
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tgcatgcctg cagggtccgaa tttctgccat tcatccgctt attatcactt attcaggcgt 120  
agcaccaggc gtttaagggc accaataact gccttaaaaa aattacgccc cgccctgcca 180  
ctcatcgag tactgttgta attcattaag cattctgccg acatggaagc catcacaac 240  
ggcatgatga acctgaatcg ccagcggcat cagcaccttg tcgccttgcg tataatattt 300  
gccccatggtg aaaacggggg cgaagaagtt gtccatattg gccacgttta aatcaaaact 360  
ggtgaaactc acccagggat tggctgagac gaaaaacata ttctcaataa acccttttagg 420  
gaaataggcc aggttttcac cgtaacacgc cacatcttgc gaatatatgt gtagaaactg 480  
ccggaaatcg tcgtggtatt cactccagag cgatgaaaac gtttcagttt gtcattggaa 540  
aacggtgtaa caagggtgaa cactatccca tatcaccagc tcaccgtctt tcattgccat 600  
acggaattcc ggatgagcat tcatcaggcg ggcaagaatg tgaataaagg ccggataaaa 660  
cttgctgctta tttttcttta cggctcttta aaaggccgta atatccagct gaacgggtctg 720  
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ggatatatca acggtggtat atccagtgat ttttttctcc atttttagctt ccttagctcc	840
tgaaaatctc gataactcaa aaaatacgcc cggtagtgat cttatttcat tatggtgaaa	900
gttggaacct cttacgtgcc gatcaacgtc tcattttcgc caaaagttgg cccagggctt	960
cccggtatca acagggacac caggatttat ttattctgcg aagtgatctt ccgtcacagg	1020
tatttattcg gactctagat aacttcgtat agcatacatt atacgaagtt atggatcatg	1080
gctgtgcagg tcgtaaatca ctgcataatt cgtgtcgctc aaggcgact cccgttctgg	1140
ataatgtttt ttgcgccgac atcataacgg ttctggcaaa tattctgaaa tgagctgttg	1200
acaattaatc atccggctcg tataatgtgt ggaattgtga gcggataaca atttcacaca	1260
ggaaacagac	1270

<210> 12

<211> 30

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer GalB1

<400> 12	30
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<210> 13

<211> 28

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer GalC11

<400> 13	28
agaaagataa gcaccgagga tcccgata	

<210> 14

<211> 26

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer GlkB1

<400> 14  
 aacaggagtg ccaaacagtg cgccga 26

<210> 15  
 <211> 30  
 <212> DNA  
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<220>  
 <223> Primer G1kC11

<400> 15  
 ctattcggcg caaaatcaac gtgaccgcct 30

<210> 16  
 <211> 99  
 <212> DNA  
 <213> Artificial Sequence

<220>  
 <223> Primer edd1

<400> 16  
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 tctgcttatc tcgcccggat ttatcgataa gctggatcc 99

<210> 17  
 <211> 98  
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<220>  
 <223> Primer edd2

<400> 17  
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 gaataattca cgtcctgtcg gatgcatatg gcggccgc 98

<210> 18  
 <211> 22  
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 <213> Artificial Sequence

<220>

<223> Primer edd3

<400> 18  
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<210> 19

<211> 21

<212> DNA

<213> Artificial Sequence

  

<220>

<223> Primer edd4

<400> 19  
actgcacact cggtacgcag a 21

<210> 20

<211> 29

<212> DNA

<213> Artificial Sequence

  

<220>

<223> CN1, encoding mutated trc promoter driving glk expression

<400> 20  
ctgacaatta atcatccggc tcgtataat 29

<210> 21

<211> 29

<212> DNA

<213> Artificial Sequence

  

<220>

<223> CN2, encoding parent trc promoter

<400> 21  
ttgacaatta atcatccggc tcgtataat 29

<210> 22

<211> 25



<212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> Primer gapA1  
 <400> 22  
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 <210> 23  
 <211> 25  
 <212> DNA  
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 <223> Primer gapA2  
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 <210> 24  
 <211> 35  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> Primer gapA3  
 <400> 24  
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 <210> 25  
 <211> 35  
 <212> DNA  
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 <220>  
 <223> Primer gapA4  
 <400> 25  
 agctggtgga atatgtgact atcaaagtag gtatc 35  
  
 <210> 26

<211> 35  
 <212> DNA  
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 <220>  
 <223> Primer gapA5  
 <400> 26  
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 <210> 27  
 <211> 35  
 <212> DNA  
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 <220>  
 <223> Primer gapA6  
 <400> 27  
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 <210> 28  
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 <223> short 1.5 GI promoter  
 <400> 28  
 gcccttgact atgccacatc ctgagcaaat aattcaacca ct 42  
  
 <210> 29  
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 <223> Primer gapA-R1  
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atgtggcata gtcaagggca tatgaatatc ctccttag 98

<210> 30

<211> 80

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer gapA-R2

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tgtaggctgg agctgcttcg 80

<210> 31

<211> 42

<212> DNA

<213> Artificial Sequence

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<223> short 1.20 GI promoter

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<210> 32

<211> 42

<212> DNA

<213> Artificial Sequence

<220>

<223> short 1.6 GI promoter

<400> 32

gcccttgaca atgccacatc ctgagcaaat aattcaacca ct 42

<210> 33

<211> 24

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer gapA-R3

<400> 33  
gtcgacaaac gctggtatac ctca 24

<210> 34

<211> 98

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer gapA-R4

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agtcatatat tccaccagct atttgtagt gaataaaagt ggttgaatta tttgctcagg 60  
atgtggcatt gtcaagggca tatgaatatt ctccttag 98

<210> 35

<211> 98

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer gapA-R5

<400> 35  
agtcatatat tccaccagct atttgtagt gaataaaagt ggttgaatta tttgctcagg 60  
atgtggcatt gtcaagggca tatgaatatt ctccttag 98

<210> 36

<211> 60

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer mgsA-1

<400> 36  
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<210> 37

<211> 60  
 <212> DNA  
 <213> Artificial Sequence  
  
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 <223> Primer mgsA-2  
 <400> 37  
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 <210> 38  
 <211> 22  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> Primer mgsA-3  
 <400> 38  
 cttgaattgt tggatggcga tg 22  
  
 <210> 39  
 <211> 21  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> Primer mgsA-4  
 <400> 39  
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 <210> 40  
 <211> 100  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <223> Primer PppcF  
 <400> 40  
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aattcctgct atttattcgt gtgtaggctg gagctgcttc 100

<210> 41

<211> 100

<212> DNA

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<220>

<223> Primer Pppcr

<400> 41

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ggatgtggca ttgtcaaggg catatgaata tcctccttag 100

<210> 42

<211> 30

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer Seqppcr 7

<400> 42

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<210> 43

<211> 90

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer 3G144

<400> 43

ccaggctgat tgaaatgccc ttctgtttca ggcataaagc cccaaagtca taaagtacac 60

tggcagcgcg gtgtaggctg gagctgcttc 90

<210> 44

<211> 93

<212> DNA

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<223> Primer 3G145

<400> 44

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cattgtcaag ggcattccgg ggatccgtcg acc 93

<210> 45

<211> 25

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer YCIKUp

<400> 45

gataataaccg cgttcatcct gggcc 25

<210> 46

<211> 25

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer YCIKDn

<400> 46

gcgagttcac ttcattgggcg tccat 25

<210> 47

<211> 98

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer yqhCFRTF

<400> 47

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acgtttaaac tcacggctgt gtaggctgga gctgcttc 98

<210> 48

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<223> Primer cyqhD1.6

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<210> 49  
<211> 46  
<212> DNA  
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<223> Primer yqhBF

<400> 49  
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<210> 50  
<211> 100  
<212> DNA  
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<223> Primer pta 1

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atcgatgcag taaatgggtga tgtgtaggct ggagctgctt 100

<210> 51  
<211> 100  
<212> DNA  
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<223> Primer ack-pta 2

<400> 51  
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cagtgcgcca cgggacaggt catatgaata tcctccttag 100

<210> 52

<211> 20

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer ack-U

<400> 52  
attcattgag tcgtcaaatt 20

<210> 53

<211> 20

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer ack-D

<400> 53  
attgcggaca tagcgcaaatt 20

<210> 54

<211> 98

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer ptSHFRT1

<400> 54  
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<210> 55

<211> 97

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer crrFRT11

<400> 55  
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<210> 56

<211> 36

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer crrR

<400> 56  
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<210> 57

<211> 100

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer DalDAF

<400> 57  
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<210> 58

<211> 100

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer DalDAR

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tgcgccgccca ataccggaatt catatgaata tcctccttag	100
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<223> Primer aldAF300	
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<223> Primer aldAR300	
<400> 60	20
gaaaaaagtg actgccgaag	
<210> 61	
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<212> DNA	
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<223> Primer DalDBF	
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<223> Primer DalDBR

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<210> 63

<211> 22

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<223> Primer alDBF

<400> 63  
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<210> 64

<211> 21

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<223> Primer alDBR

<400> 64  
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<210> 65

<211> 13669

<212> DNA

<213> Artificial sequence

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<223> Plasmid pSYCO101

<400> 65  
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acgcttaaaa ataataaaaag cagacttgac ctgatagttt ggctgtgagc aattatgtgc 180  
ttagtgcac taacgcttga gttaagccgc gccgcgaagc ggcgtcggct tgaacgaatt 240

gttagacatt atttgccgac taccttggtg atctcgccctt tcacgtagtg gacaaattct	300
tccaactgat ctgcgcgga ggccaagcga tcttcttctt gtccaagata agcctgtcta	360
gcttcaagta tgacgggctg atactgggcc ggcaggcgct ccattgcca gtcggcagcg	420
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actacatttc gctcatcgcc agcccagtcg ggcggcgagt tccatagcgt taaggtttca	540
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